

Fig. 1A.

Replacement Sheet

M V N R S V A F S A F V L I L F V L A I
1 ATGGTAATCGGTCGGTTCCTCCGCCGTTCTGATCCTTTCGTGCTCGGCATC
S
61 TCAGGTATCAATCTTAGTTCATTATTGAATAGTAGTATTATATCTTTATGG
Intron
121 TTTTATGTGTTCTGACAGTTGCCAATATTGAGTAGATATCGCATCCGTTAGTGAGAAC
D I A S V S G E
L C E K A S K T W S G N C G N T G H C D
181 TATGCCGAGAAAGCTAGCAAGACATGTCGGGAAACTGTGCCAATACGGGACATTGTGACA
N C O I
241 ACCAATGTAATCATGGAGGGTGGCCCATGAGCGTGCATGTGCGTAACGGGAAC
HindIII
301 H M C F C Y F N C K K A E K L A Q D K L
ACATGTGTTCTGTTACTTCAATTGTA AAAAAGCCGAAAGCTTGCTCAAGACAACTTA
HindIII
361 K A E Q L A Q D K L N A Q K L D R D A K
AAGCCGAACTCGCTCAAGACAACTTAATGCCCAAAGCTTGACCGTGATGCCAAGA
K V V P N V E H P
421 AAGTGTTCCAAACGTTGAACATCCG



Fig.1B.

Replacement Sheet

1	GTCCCCGGGTCACGAAGTTCGGCACATCTTAGCGTTATGCATAAGTCAAAATGGCCAA	M A K
61	AAATTCAGTTGCTTCTTGCATTGTGCCTGCTTCTTTCATTCTTGCTATCTCAGAAAT	N S V A F E A L C L L L F I L A I S E I
121	CAGATCGGTGAAGGGGAATTATGTGAGAAGGCAAGCAAGACATGCTCTGGAATTGTGG	R S V K G E L C E K A S K T W S G N C G
181	CAATACAAGACACTGTGATGACCAGTGCAAGTCTTGGAGGGTGCAGCCCATGAGCTTG	N T R H C D D Q C K S W E G A A H G A C
241	TCACGTGCGCGGTGGGAACACATGTGCTTCTGCTACTTCAACTGTCCCAAGCCCCAGAA	H V R G G K H M C F C Y F N C P K A Q K
301	GTGGCTGAGGATAAATCTCAGAGCAGCAGAGCTAGCAAGAGAAGATAATATTGGAGC	L A E D K L R A A E L A K E K N N I G A
361	TGAAAAGGTGCCCTTCAGCCACACCTTGAGTACTAACAAA	E K V P S A T P

Fig.2A.

M A K N S V A F L A F L L L L F V
1 GGCACGAGTAATGGCCAAAATTCAGTGCCTTCTTAGCATTTCTTCTGCTTCTTTTCGT
L A I S E I G S V K G E L C E K A S K T
61 TCTTGCTATCTCAGAAATCGGATCGGTGAAGGGGAATTATGTGAGAAGGCAAGCAAGAC
W S G N C G N T R H C D D Q C K S W E G
121 ATGGTCTGGAATTGTGGCAATACAAGACACTGTGATGACCAGTGCAAGTCTTGGGAGGG
A A H G A C H V R G G K H M C F C Y F N
181 CGCAGCCCATGGAGCTTGTACGTGCGCGGTGGAAACACACATGTGCTTTTGCTACTTCAA
C S K A Q K L A Q D K L K A D K L A K E
241 CTGTTCCAAAGCCCGAAGCTGGCTCAGGATAAACTCAAAGCCGACAAGCTCGCCAAGGA
K S E A E K V P A T P
301 GAAGAGTGAAGCCGAAAAGGTGCCAGCTACACCTTGAGTACTAACAAGTGTGTATGATT
361 ATGAATAAAGAGAAAATGCTTTCTAGTTACCATATTTAGCATTTCTCTAATGTGTAATGTT
421 TGTTGCTTTTGGAACTAATTGCCTTAACATATGATCCAGCTAATAATGTTTAAGTATATA
481 ATATAAGTTATCTTATTTTGAAGCCCTGTAAAAAATAAAAAA

Fig.2B.

1 CGGCACGAGGCACATCTCAAAAATGGCCAAAATTCGGTTGCTTCTTGCAATTGTCC
| M A K N S V A F F A F V
61 L L L F V L A I S E I G S V K G E L C E
TGCTTCTTTTCGTTCTTGTCTATCTCAGAAATTGGATCGGTGAAGGAGAATTATGTGAGA
K A S K T W S G N C G I T S H C D N Q C
121 AGGCAAGCAAGACATGGTCTGGAATTGTGGCATCACATCACA CTGTGACAACCA GTGCC
R S W E G A I H G A C H V R G G K H M C
181 GGTGCGGAGGGTGCAATCCATGAGCTTGTACGTGCGCGGTGGGAACACATGTGCT
F C Y F N C S K A D E L A K E K I E A E
241 TCTGCTACTTCAACTGTTCCAAAGCCGATGAGCTCGCGAAGGAGAAGATTGAAGCCGAAA
K M P A T P
301 AGATGCCACGCCACACCTTGAGTACTAACAATAATGCTATATGATTATAATAAGAGAAAAT
361 GCTTCTAAAAA AAAAAAAAAA

Fig.3.

1 GGCACGAGCCCTATTAAAAAATGCTGAATCGATCGGTGCTTCTCCGTGTCGTTCTGAT
M V N R S V A F S V F V L I
L F V L A I S D I T S V R G E V C E K A
61 CCTTTTCGTCGCTCG[↓]CCATCTCAGATATCACAAAGTGTGAGAGGAGAAGTATCGGAGAAAGC
S K T W S G N C G N T G H C D N Q C K Y
121 TAGCAAGACATGGTCAGGAAACTGTGGCAACACGGGACACTGTGACAACCAATGTAATA
W E G A A H G A C H V R G G K H M C F C
181 CTGGAGGGGGGGCCCATGGGGCGTGCCACCGTGCGTGAGGGAACACATGTGTTCTG
Y F K C P K A E K L A Q D K V N A Q E L
241 CTACTTCAAGTGTCC[↓]AAAGCCGAAAGCTTGCTCAAGACAAAGTTAATGCCCAAGAGCT
D R D A K K V I P N V E H P
301 TGACCGTGATGCCAAGAAAGTGATTCGGAACGTGAACATCCGTGAAGGGTCGGTTCT
361 TTAATAGAAAGTCTTAGATTACGAATCGGAATACTATAGAAAATGTTGCTAAATGTC
421 ACATTATAATTAGAACTTTATGATTGTTGTCAATAGGGCATTCTTCTGTTAGTGATATGT
481 GTAATAAGGTGATGCTTTTATGCTTTTCGTGCGTAAGAGTTTTCGACTATGTGTAATAAA
541 GAAAGGGTCTTTT TTTT TAAAAA AAAAAA AAAAAA A

Fig.4.

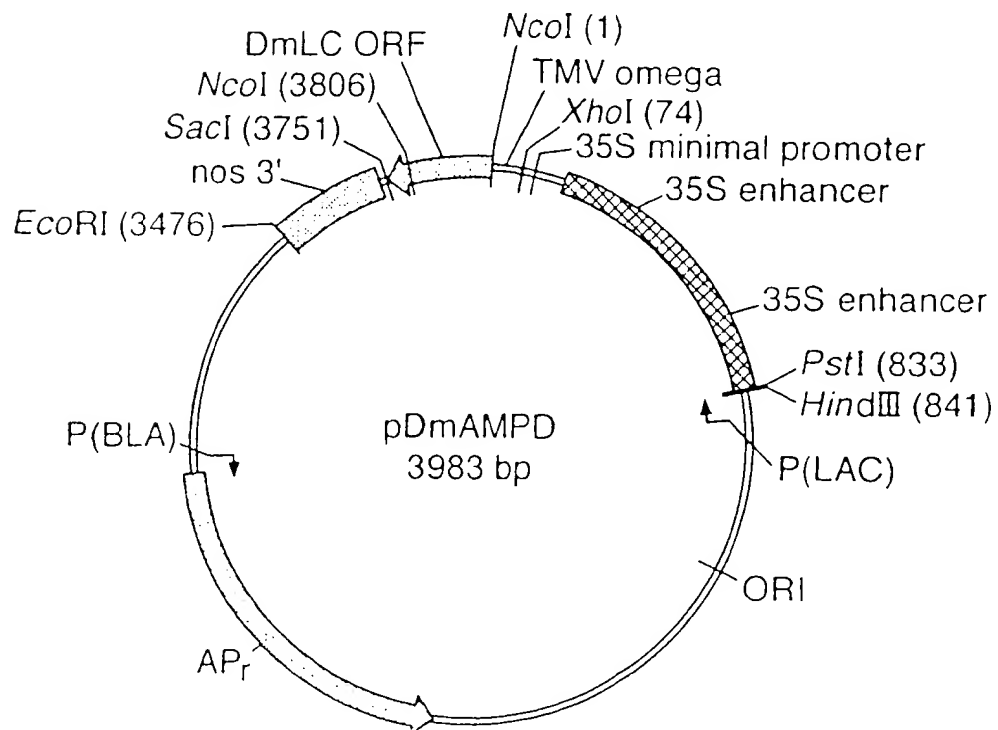
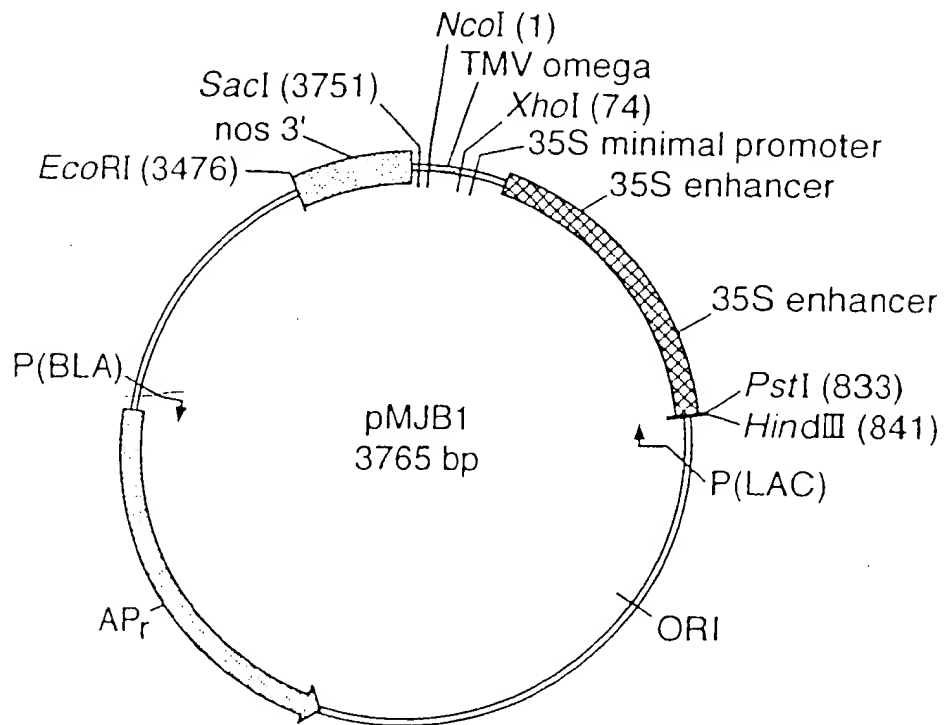


Fig.4 (Cont).

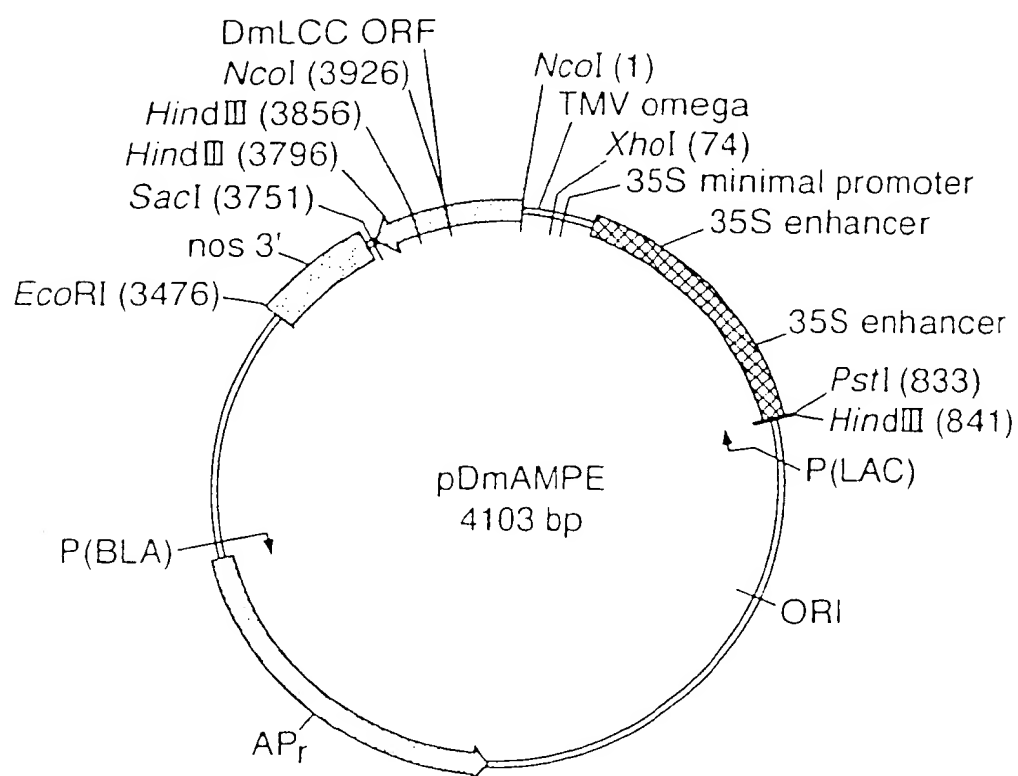
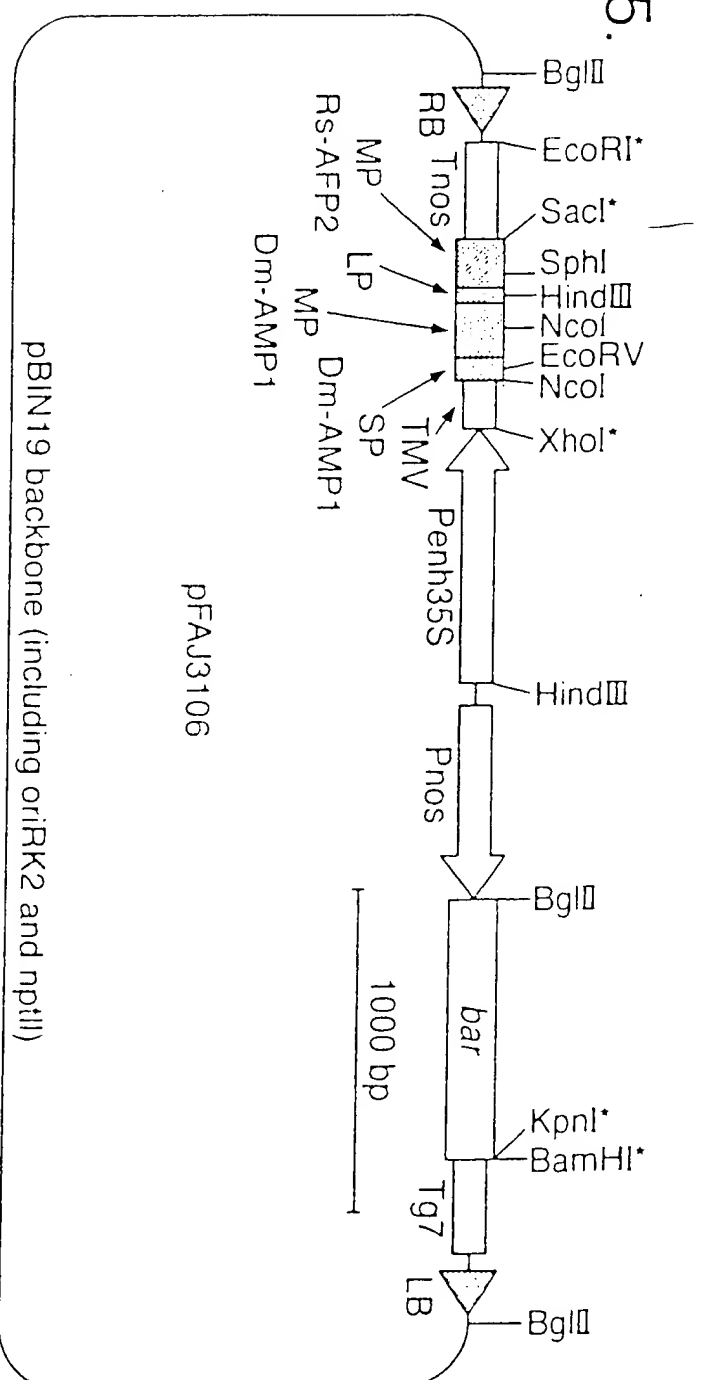


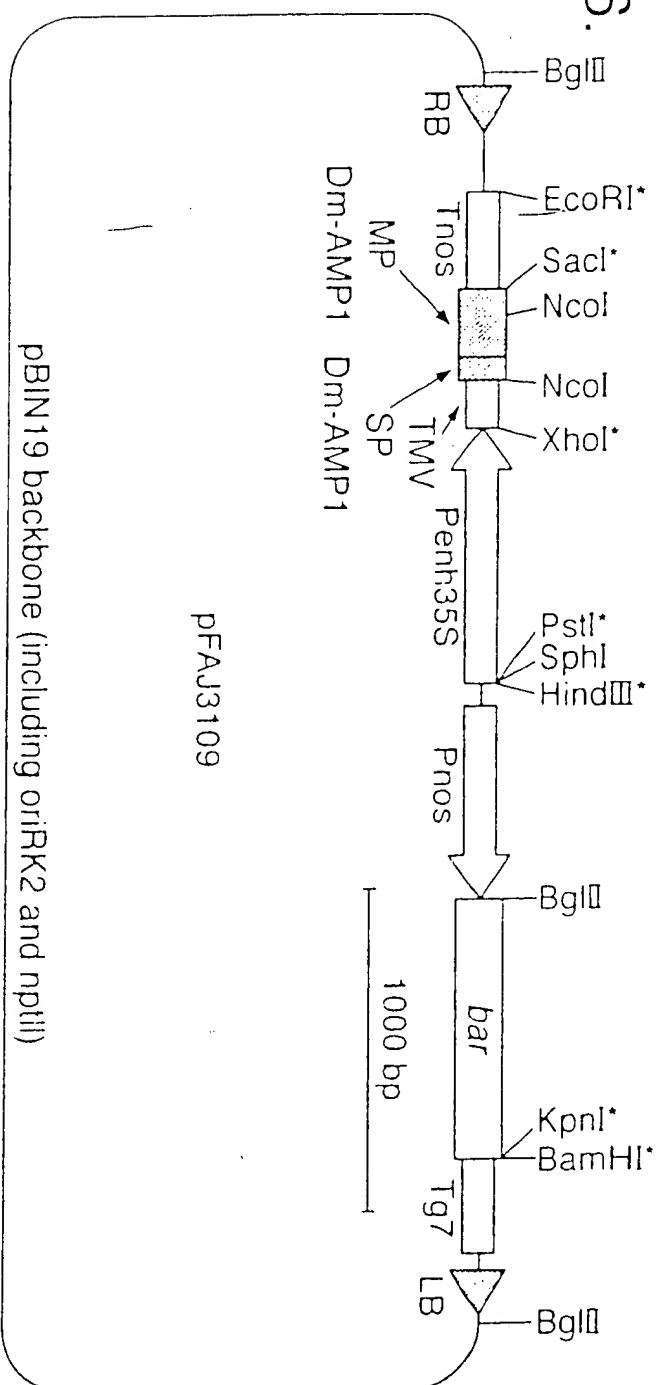
Fig. 5.



Symbols

- RB: right border of T-DNA
- Tnos: terminator of T-DNA nopaline synthase gene
- MP Rs-AFP2: mature protein domain of Rs-AFP2
- LP: first 16 AA of Dm-AMP1 C-terminal propeptide and subtilisin-like protease recognition site IGKR
- MP Dm-AMP1: mature protein domain of Dm-AMP1 cDNA
- SP Dm-AMP1: signal peptide domain of Dm-AMP1 cDNA
- TMV: tobacco mosaic virus 5' leader sequence
- Penh35S: promoter of 35S RNA of cauliflower mosaic virus with duplicated enhancer region
- Pnos: promoter of T-DNA nopaline synthase gene
- bar: basta resistance encoding gene
- Tg7: terminator of T-DNA gene 7
- LB: left border of T-DNA
- *: unique restriction site

Fig.6.



Symbols

- RB: right border of T-DNA
- Tnos: terminator of T-DNA nopaline synthase gene
- MP Dm-AMP1: mature protein domain of Dm-AMP1
- SP Dm-AMP1: signal peptide domain of Dm-AMP1 cDNA
- TMV: tobacco mosaic virus 5' leader sequence
- Penh35S: promotor of 35S RNA of cauliflower mosaic virus with duplicated enhancer region
- Pnos: promotor of T-DNA nopaline synthase gene
- bar: basta resistance encoding gene
- Tg7: terminator of T-DNA gene 7
- LB: left border of T-DNA
- *: unique restriction site

Fig.7.

PFAJ3106

XhoI

CTCGAGTATTTTACACAATTACCAACAACAACAACAACAACAATTACAATTACT

NCOI

ATTTACAATTACACCATGGTGAATCGGTGCGTTGCGTTCGCCGTTCTGATCCTT
M V N R S V A F S A F V L I L

TTGTCCTCGCCATCTCAGATATCGCATCCGTTAGTGAGAACTATGCGAGAAAGCTAGC
F V L A I S D I A S V S G E L C E K A S

AAGACGTGTCGGGCAACTGTGGCAACACGGGACATTGTGACAACCAATGTAATCATGG
K T W S G N C G N T G H C D N Q C K S W

GAGGTCGCCCATGAGCGTGTCTATGTGCCCTAACGGGAACACATGTGTTCTGTTAC
E G A A | H G A C H V R N G K H M C F C Y

TTCAATTGTA AAAAAGCCGAAAAGCTTGCTCAAGACA AACTTAAGCCGAACA CTCA TC
F N C K K A E K L A Q D K L K A E Q L I

GGAAGAGGCAGAACTGTGCCAAAGCCCAAGTGGACATGCTCAGAGTCTGTGGAAC
G K R Q K L C Q R P S G T W S G V C G N

AATAACGCATGCAAGAATCAGTGCATTAGACTTGAGAAAAGCAGACATGATCTTGCAAC
N N A C K N Q C I R L E K A R H G S C N

TATGTCCTTCCAGCTCACAAGTGATCTGCTACTTTCCTTGTTAATAGGAGCTC
Y V F P A H K C I C Y F P C - - SacI

pFAJ3109

Fig.8.

XhoI

CTCGAGTATTTTACACAATTACCACAACAACAACAACAACATTACAATTACT

NcoI

ATTTACAATTACACCATGGTGAATCGGTCGGTTGCCGTTCTCCGGCTTCTGATCCTT
M V N R S V A F S A F V L I L

TTTCGTCTCGCCATCTCAGATATCCGATCCGTTAGTGAGAACTATGCGAGAAAGCTAGC
F V L A I S D I A S V S G E L C E K A S

AAGACGTGTCGGGCACTGTGCCAACACGGGACATTGTGACAACCAATGTAATCATGG
K T W S G N C G N T G H C D N Q C K S W

GAGGTCGGCCCATGAGCGTGTCAATGTGCGTAATGGAAACACATGTGTTCTGTAC
E G A A H G A C H V R N G K H M C F C Y

SacI

TTCAATTGTTGAGCTC
F N C -

Fig.9.

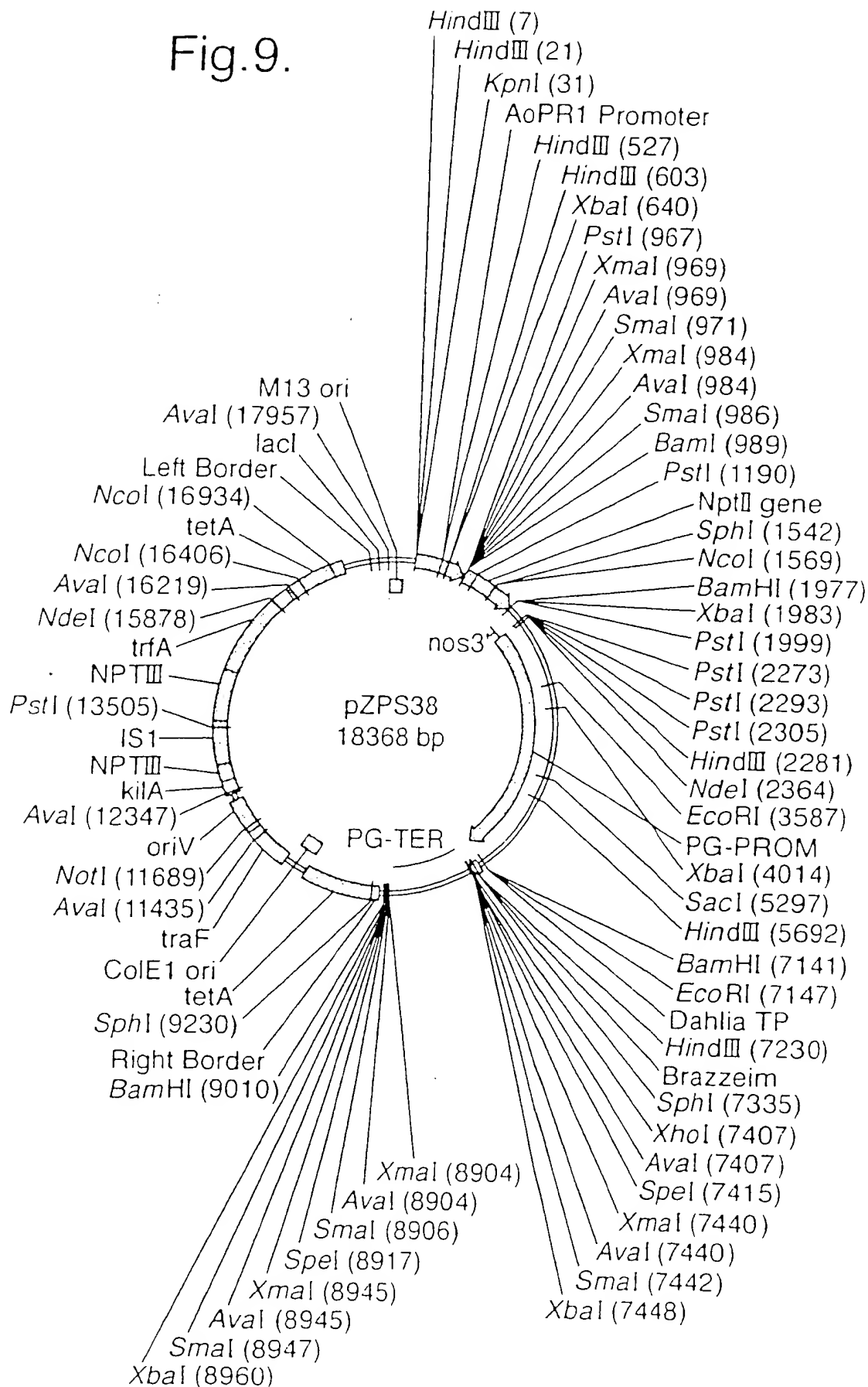


Fig.10.

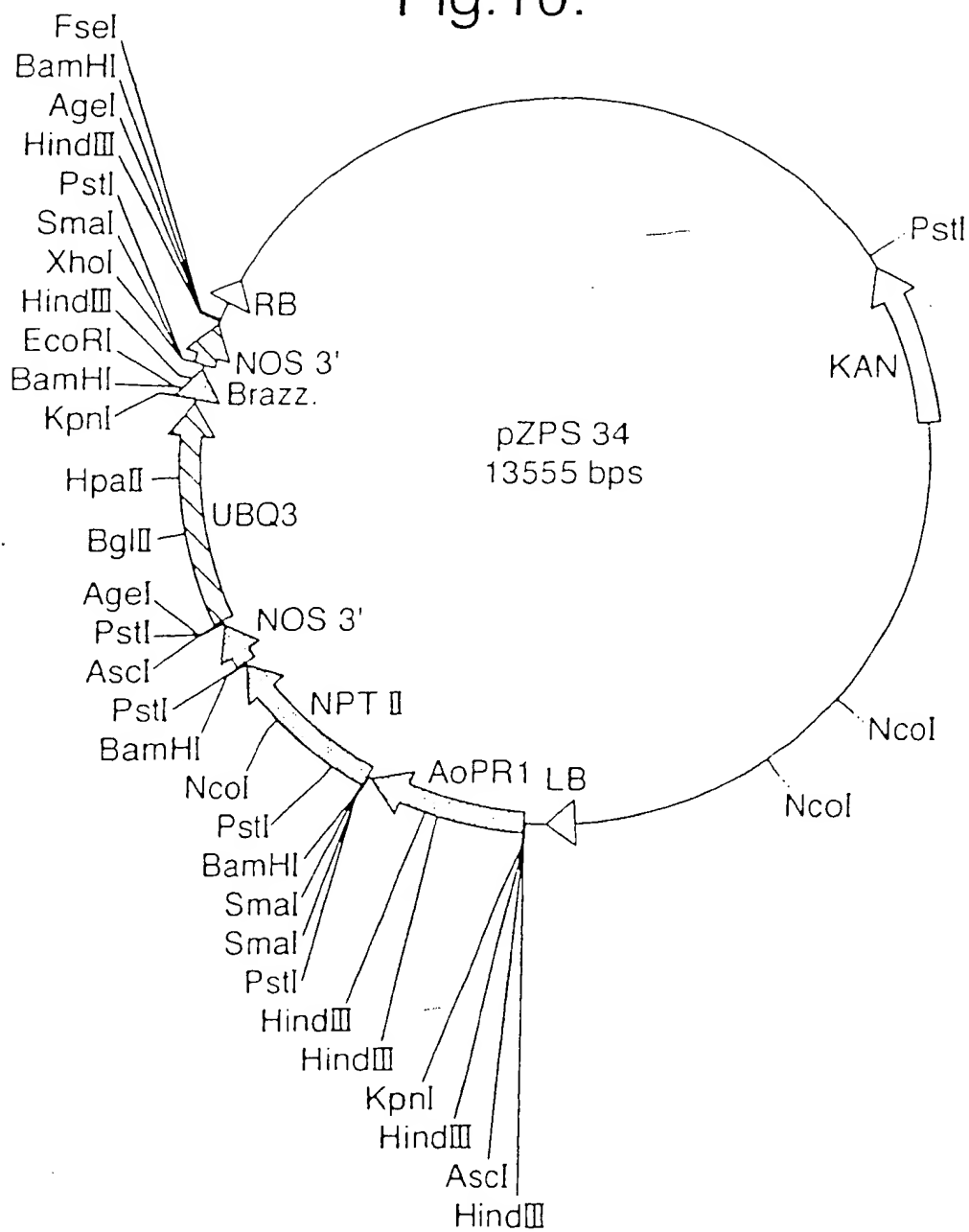


Fig.11.

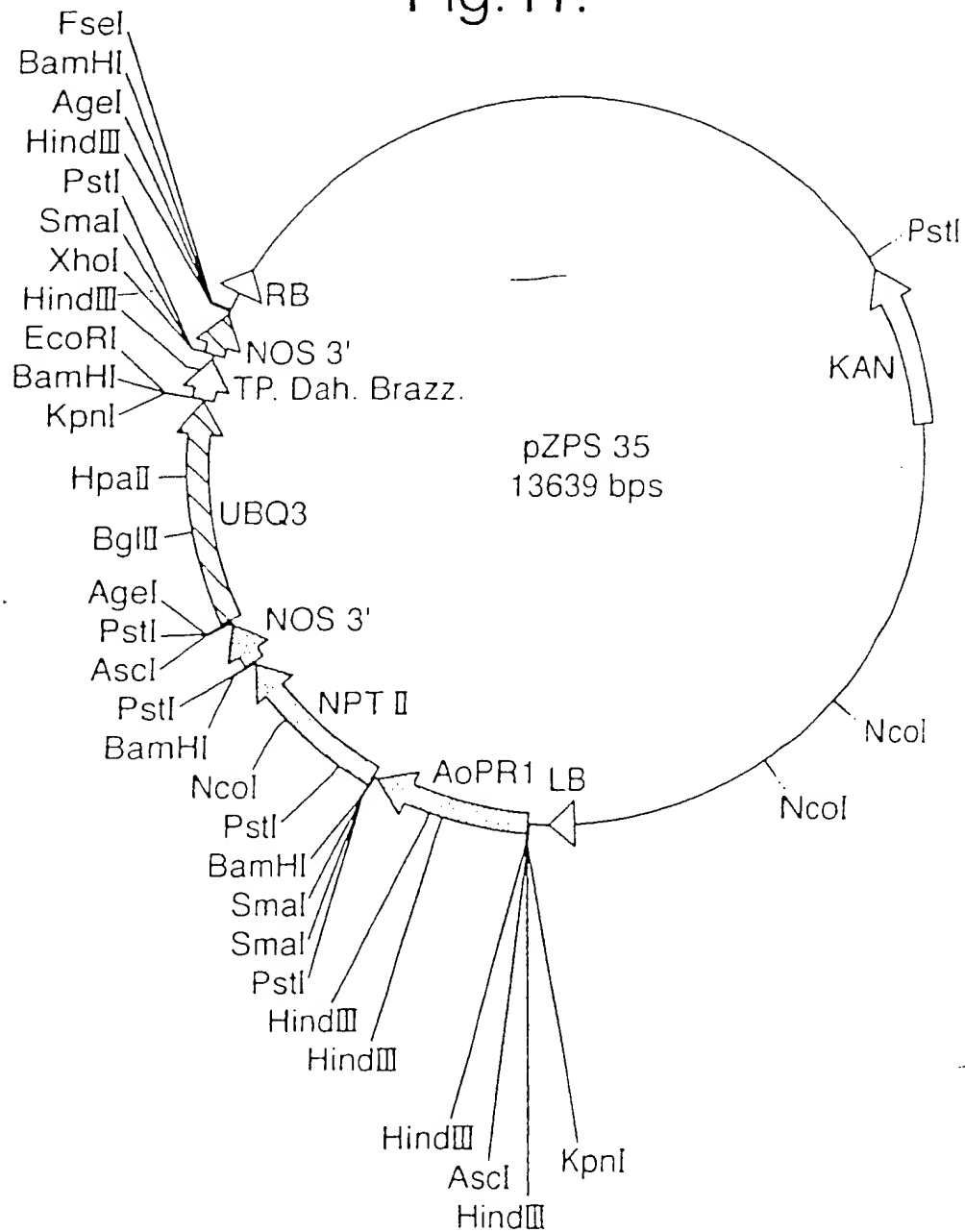


Fig.12.

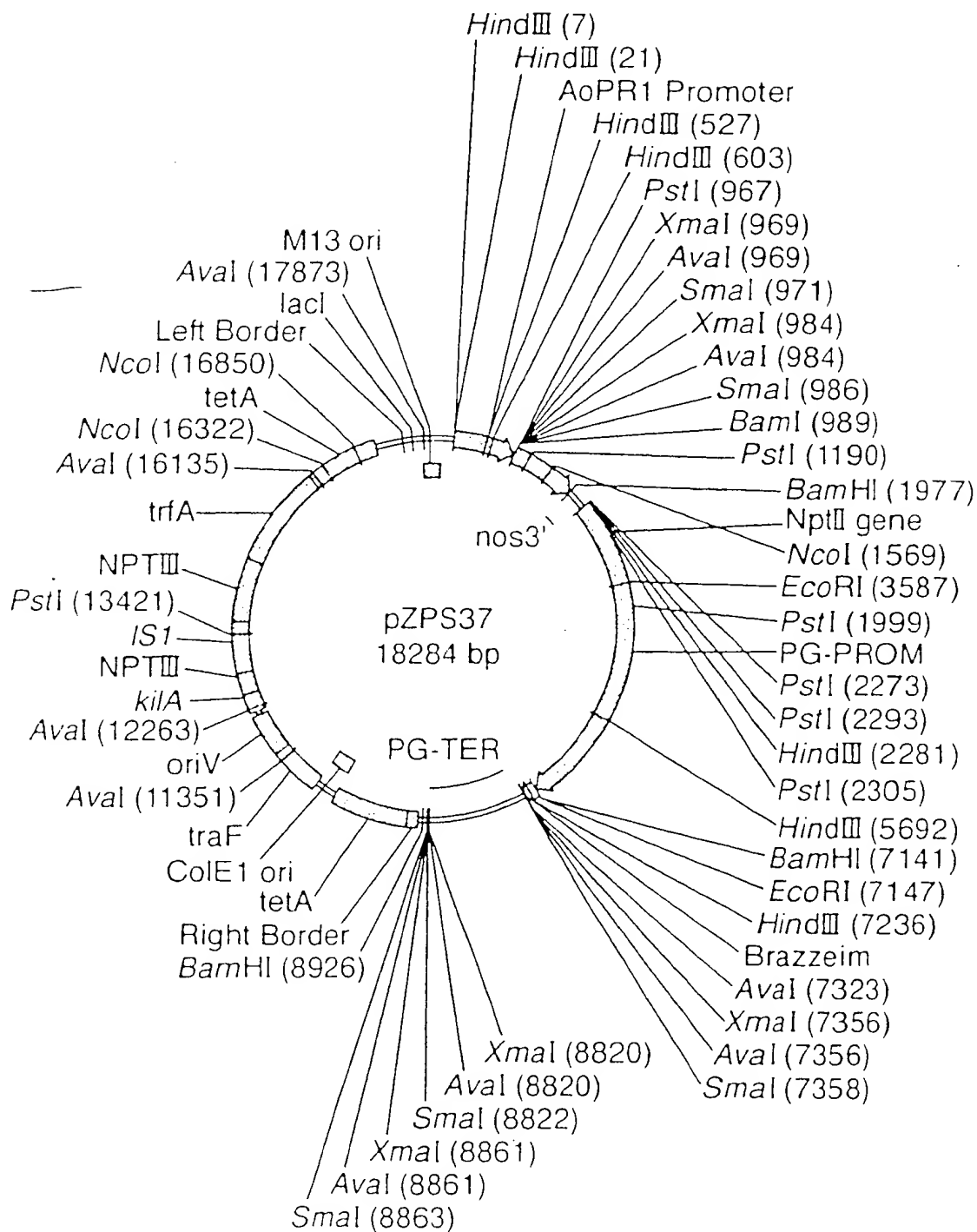
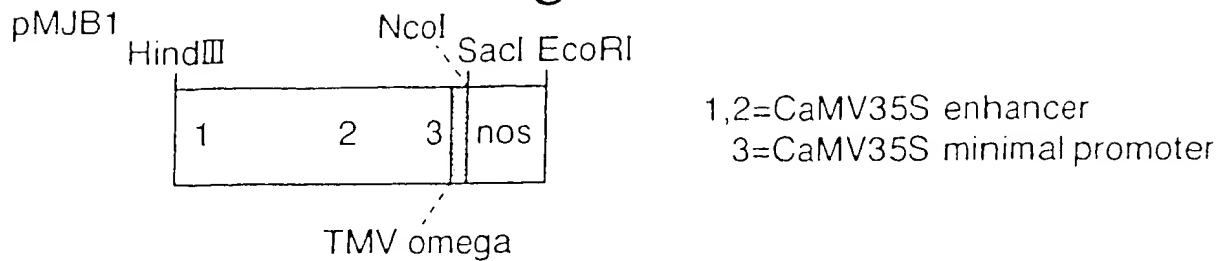
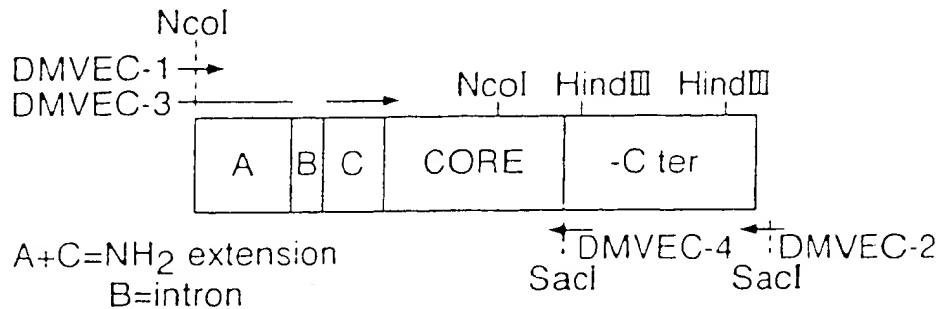


Fig.13.



Structure of DmAMP1 Gene and position of vector construction oligonucleotides

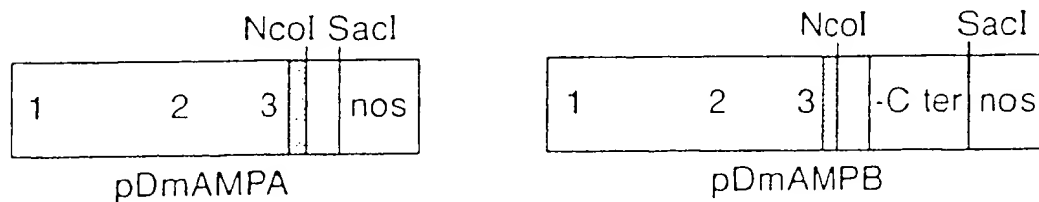


PCR Dahlia genomic DNA with DMVEC-1 and DMVEC-2, isolate 450 bp product.

PCR 450 bp DMVEC-1/DMVEC-2 PCR product with DMVEC 1 and 4.

Isolate 60 bp NcoI / SacI fragment, clone into pMJB1 NcoI / SacI=pDmAMPA.

Cut 450 bp DMVEC-1/DMVEC-2 PCR product NcoI / SacI. Isolate 180 bp NcoI / SacI fragment, clone into pMJB1 NcoI / SacI =pDmAMPB



PCR 450 bp DMVEC-1/DMVEC-2 PCR product with DMVEC 3 and 4.

Isolate 150 bp NcoI fragment, clone into pDmAMPA

and pDmAMPB NcoI =pDmAMPD and pDmAMPE

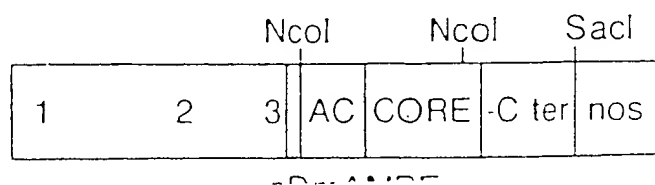
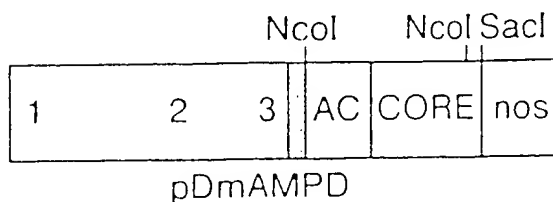


Fig.14.

Sequence	
ID No. 6	Dm-AMP1
	GAG CTT TGC GAG AAG GCT TCT AAG ACT TGG TCT GGA AAC
	TGG GAG GGA GCT GCT CAT GGA GCT TGC CAT GTT AGA AAC
Sequence	
ID No. 7	Dm-AMP2
	GAG GTT TGC GAG AAG GCT TCT AAG ACT TGG TCT GGA AAC

| Fig.14 (Cont).

TGC GGA AAC ACT GGA CAT TGC GAT AAC CAA TGC AAG TCT

GGA AAG CAT ATG TGC TTC TGC TAC TTC AAC TGC

TGC GGA AAC ACT GGA CAT TGC

|

Fig.15.

